ISTANBUL AYDIN UNIVERSITY
ENGINEERING FACULTY
INTERNATIONAL JOURNAL OF FOOD ENGINEERING RESEARCH
(IJFER)

Year **7** Issue **2** - **October 2021**

International Journal of Food Engineering Research (IJFER) ISSN: **2149-5777**

October **2021** Year: **7** Number: **2**

Language
English

Publication Period
Published twice a year
April and October

http://ijfer.aydin.edu.tr/tr/editorler-kurulu/

Correspondence Address
İstanbul Aydın University, Faculty of Engineering
Department of Food Engineering
Florya Yerleşkesi, İnönü Caddesi, No. 38, Küçükçekmece, İstanbul,
Turkey Phone: +90 212 411 61 00
Fax: +90 212 425 57 59

Printed by
Levent Baskı Merkezi - **Sertifika No:** 35983
Adres: Emniyetevler Mahallesi Yeniçeri Sokak No:6/A
4. Levent / İstanbul, Türkiye
Tel: 0212 270 80 70
E-mail: info@leventbaskimerkezi.com

International Journal of Food Engineering Research (IJFER) is peer-reviewed journal which provides a platform for publication of original scientific research and applied practice studies. Positioned as a vehicle for academics and practitioners to share field research, the journal aims to appeal to both researchers and academicians.

Aims and Scope

International Journal of Food Engineering Research (IJFER) is an international, peer-reviewed journal devoted to the publication of high quality original studies and reviews concerning a broad and comprehensive view of fundamental and applied research in food science&technology and their related subjects as nutrition, agriculture, food safety, food originated diseases and economic aspects.

IJFER is an international periodical published twice a year (April and October). The journal is published in both print and electronic format.

From The Editor

International Journal of Food Engineering Research (IJFER) has been publishing by Istanbul Aydın University Faculty of Engineering Department of Food Engineering since 2015. The journal covers wide ranges of area such as Food Processing, Food Preservation, Food Microbiology, Food Chemistry, Biotechnology, Nanotechnology, Novel Technologies, Food Safety, Food Security, Food Quality and their related subjects as nutrition, food and health, agriculture, economic aspects and sustainability in food production.

Food Engineering is getting more and more attention because it is directly related to human health. While the food and drinks we eat help to protect our health, on the other hand, improper conditions during the conversion of the raw material to the product, the use of poor quality raw materials, and the employees not working under hygienic conditions can cause the food harmful to health. Our aim in this journal is to include the recent research and reviews on food and beverages from field to fork. Articles submitted to the journal are accepted for publication after being reviewed by expert referees.

In the following years, the journal will include scientific activities such as symposiums, congresses, conferences and workshops held in the field of food science and technology, and information about the books published in this field. We hope that the journal will be a good resource for engineers, experts, researchers and students working in the food industry.

Prof. Dr. Z. Dilek Heperkan
Editor

International Journal of Food Engineering Research (IJFER)

Contents

Research Articles

Review Articles

Review Articles

DOI Numbers

Genel DOI: 10.17932/IAU.IJFER.2015.003
IJFER Ekim 2021 Cilt 2 Sayı 1 DOI: 10.17932/IAU.IJFER.2015.003/2021.702

PRODUCTION OF COLD GREEN TEA WITH NATURAL ADDITIVES

Atilla Oktamış Tanyıldız, Elif Çulhacı, Meral Yıldırım Yalçın*

[1]*Department of Food Engineering, Istanbul Aydın University, 34295, Istanbul, Türkiye*
*Corresponding author: *meralyildirimyalcin@aydin.edu.tr*
ORCID number: 0000-0002-5885-8849

ABSTRACT

Tea is the product that goes through production stages such as withering, rolling, shredding, oxidation and drying after the harvest of 2.5 leaves of the *Camellia sinensis* species. Green tea, known for its health benefits, is consumed less than black tea due to sensory features. Cold tea is a product that is widely used in the world. The cold teas on the market are made with black tea extract and contain various additives to improve its sensory properties. In this study, it was aimed to increase the nutritional and sensory properties of cold green tea by adding natural substances: honey and lemon juice, and peel. Also, the effects of brewing methods on the quality of tea were investigated. Green tea samples containing lemon peel were infused by heat-assisted method and ultrasound method. Cold tea samples were produced with different concentrations of honey and lemon juice addition. The physiochemical parameters (total dry matter, pH, and color properties), total phenolic content, antioxidant activity, and sensory analysis of the cold tea samples were evaluated. The optimal tea formula determined according to the results is the sample of containing high amount of lemon juice. While the total phenolic content was 285.76 ± 2.80 mg GAE/ 1mL tea and the antioxidant activity was 62.42 ± 6.15 (DPPH radical scavenging activity, %) in the green tea sample, the total phenolic content was found to be 300.76 ± 3.34 mg GAE/ 1mL tea and the antioxidant activity 91.46 ± 1.94 (DPPH radical scavenging activity, %) in the sample containing high amount of lemon juice. It was observed that the honey and lemon juice addition increased the total phenolic and antioxidant content of the control sample. When the results of the study and the

[1]Geliş Tarihi: 04.06.2021 Kabul Tarihi: 19.07.2021
DOI: 10.17932/IAU.IJFER.2015.003/ijfer_v07i2001

sustainability approach are evaluated, a product that will meet the market needs has been obtained.

Keywords: Green tea, Honey, Lemon, Ultrasound, Antioxidant activity.

INTRODUCTION

Tea, which is stated in the sources of China to be found by chance falling into boiling water, has spread all over the world and reached many different cultures. About 30 countries, mainly India, Taiwan, and China, are involved in tea production. The presentation, consumption, and brewing of tea differ from country to country. Merkez Tea Factory started production in 1947 as Türkiye's first tea factory with a capacity of 60 tons/day. The private sector serves a daily processing capacity of approximately 9 thousand tons **[1].**

Tea polyphenols (catechins) create 30% to 42% of the dry weight of solids in brewed green tea. Flavones and flavonols have been detected in tea. Quercetin, kaempferol, myricetin, and glycosides are the main flavonols. The caffeine level is affected from the brewing time, temperature, and leaf size. Flavins are responsible for the bright red color of black tea. Thearubigins, the brown pigment in black tea, are heterogeneous polymers of tea catechins. They constitute 60% of the water-extractable material of black tea by dry weight. The dominant minerals contained in the water-soluble ash of tea are potassium, calcium, magnesium, and aluminum **[2].** Green tea is loaded with antioxidants and nutritional components; therefore, optimum brewing conditions are very important to increase the efficacy of green tea **[3].**

Epigallocatechin-3-gallate (EGCG) is a substance that has important properties such as antioxidant, anticancer, anti-inflammatory, and anti-proliferative effects and is widely used in traditional medicine. It prevents the formation of various types of cancer by inhibiting the effect of carcinogenic substances **[4].** Studies have proven that the antioxidant EGCG in tea has anti-carcinogenic effects. The effects of green tea on cardiovascular diseases are also known. It has proven antimicrobial properties against gram-positive and gram-negative bacteria. These include *Listeria*, *E. coli*, *Salmonella,* and *Pseudomonas* species. It is proven that it prevents oral wounds due to its anti-inflammatory properties **[5].**

Türkiye is the second biggest honey producer in the world after China. It has a 6.9% share in the world market **[6].** While honey consumption is common, its adulteration is widespread **[7].** Since ancient times, honey consumption was common, and its health effects were known **[8].** It is a natural bee product that has antioxidant, antimicrobial, and anti-inflammatory properties **[9].** The major components are complex of sugars and also have enzymes, minerals, vitamins, amino acids, and organic acids. Honey has low protein content (0.5%) and the proteins are mainly enzymes **[10].** The chemical composition may differ with climate, flowers, and geography **[11].** While the major components are carbohydrates, the glycemic index of honey ranges from 32 to 87 **[12].** It is known that heat treatment damages the enzymes and increases HMF (*hydroxymethylfurfural*) content in honey **[13].**

Lemon is a medicinal plant belonging to the *Rutaceae* family that grows on small trees 10 to 20 feet high. The main producers of lemon, which are preferred frequently all over the world, are Mexico, Argentina, Brazil, Spain, China, USA, Türkiye, Iran, and Italy. Extracts from other parts of lemon, which have alkaloids with anticancer activity, have been reported in various studies to have antibacterial potential. The peel of citrus fruits is rich in glycosides, coumarins, and flavonoids **[14].** Lemon peel is rich in substances such as flavonoids, pectin, fiber, and essential oil. For this reason, it is important to evaluate lemon peels in the food industry. It shows important effects such as antioxidant, antimicrobial and anti-diabetic properties. Due to the positive effects of beneficial components in lemon peels on health, it can be used in the development of functional foods **[15].**

In the traditional brewing method, the solubility of tea components is increased by the heating or boiling method **[16].** Brewing temperature and time are considered the most effective factors for the composition and sensory properties of tea infusions. Increasing temperature and time applications make the infusions dark and bitter. In addition, the antioxidant activity, the total phenolic content, and the total flavonoid contents vary depending on the brewing temperature and duration. At this point, the composition of the tea before brewing is also important. Determining the optimum brewing conditions is very important in terms of the composition and sensory properties of the prepared tea **[17].**

Ultrasonic extraction is an effective process with wide application in food processing. It is characterized by high frequency and good penetrating power **[3].** Ultrasonic waves have a frequency of 20 kHz and higher. The ultrasonic method has become increasingly popular in recent years both to prevent loss due to the negative impact of the active ingredients on the temperature and to increase the extraction yield **[16].** In ultrasonic extraction techniques, the material transfer is supported by increasing the breaking effect of the cell walls **[3].** The advantages of ultrasonic extraction compared to other methods are that the equipment cost is cheap, the process is easy, and the time saving is the case. It is stated that it will be more effective in the extraction of antioxidants, phenolics, and anthocyanins compared to other methods that require high temperatures and a long time. In addition, studies have shown that alkaloids and flavonoids can be extracted in extracts obtained from ultrasonic extraction compared to the traditional method **[18].**

The study aims to analyze the effects of different infusion methods on green tea's physicochemical properties and improve sensory acceptance by adding lemon derivatives and honey. In the study, the traditional method of high-temperature infusion and the low-temperature method with ultrasound, which is a good extraction method, were used.

MATERIALS AND METHOD

Materials

The green tea used in the preparation of cold green tea was obtained from the Çaykur (Rize, Türkiye). The added honey is the Şemdinli flower honey of the Baltek company (Istanbul, Türkiye). The lemon and water used in the study were bought from a local market in Istanbul. Lemon peels were dried at 40 ^{o}C for 8 hours in an oven (Suzen, Türkiye). Methanol (Merck, Germany), 2,2-Diphenyl-1-picrylhydrazyl (DPPH) (Sigma-Aldrıchand, USA), Folin Ciocalteu reagents (Merck, Germany), Sodium carbonate (Merck, Germany), Gallic acid anhydrous (ISOLAB chemicals, Germany) were used in the study.

Preparation of cold tea with traditional method

Since there were 3 different tea formulations and 2 different brewing methods in the study, 6 different teas were obtained: Traditionally brewed green tea (control sample) (GT), Traditionally brewed green tea with high honey content (HGT), Traditionally brewed green tea with high lemon juice content (LJGT), Green tea brewed by ultrasonic method (control sample) (UGT), Green tea with high honey content brewed by ultrasonic method (U-HGT), Green tea with high lemon juice brewed by ultrasonic method (U-LJGT).

Water (100 mL) was taken into a clean jar and placed in a water bath (Stuart SWB2D, UK) previously set at 85 ^{o}C. Green tea (2 g) was weighed for the sample named GT. Green tea (2 g) and lemon peel (0.2 g) were weighed for the sample named HGT and LJGT. All tea samples were transferred to jars in a constant temperature (85 ^{o}C) water bath and infused for 6 minutes. The samples were filtered (Sartorius Stedim Biotech, France) with the help of a strainer and left at room temperature. 9 g of honey and 1 g of lemon juice were weighed in the beaker for the HGT sample and 6 g of honey and 4 g of lemon juice for the LJGT sample. These products were added to the tea and mixed homogeneously. The prepared samples were kept in the refrigerator until the analysis time. Production steps of the traditional brewing method were demonstrated in Figure 1.

Preparation of cold tea by ultrasonic method

The same amounts of the samples by the traditional method were prepared for the ultrasound method. The brewing was carried out in an ultrasonic water bath (SWB2D, UK) at 25 ^{o}C for 30 min and outside the ultrasonic bath at 25 ^{o}C for 30 min. The stages after brewing were the same as with the traditional method. The production steps of the ultrasound proses are similar to the traditional one. The only difference is the temperature and time.

Physicochemical analyzes

Total dry matter analysis of cold tea samples was made using a drying oven (BINDER, Germany) at 105 ^{o}C. The color analysis of cold tea samples was made using Lovibond Tintometer (PFX-880, UK) according to

the CIE Lab system (L^* Lightness, a^*: redness or greenness, b^*: yellowness or blueness). The pH analysis of the samples was performed with a pH meter (Mettler Toledo, S210, Switzerland).

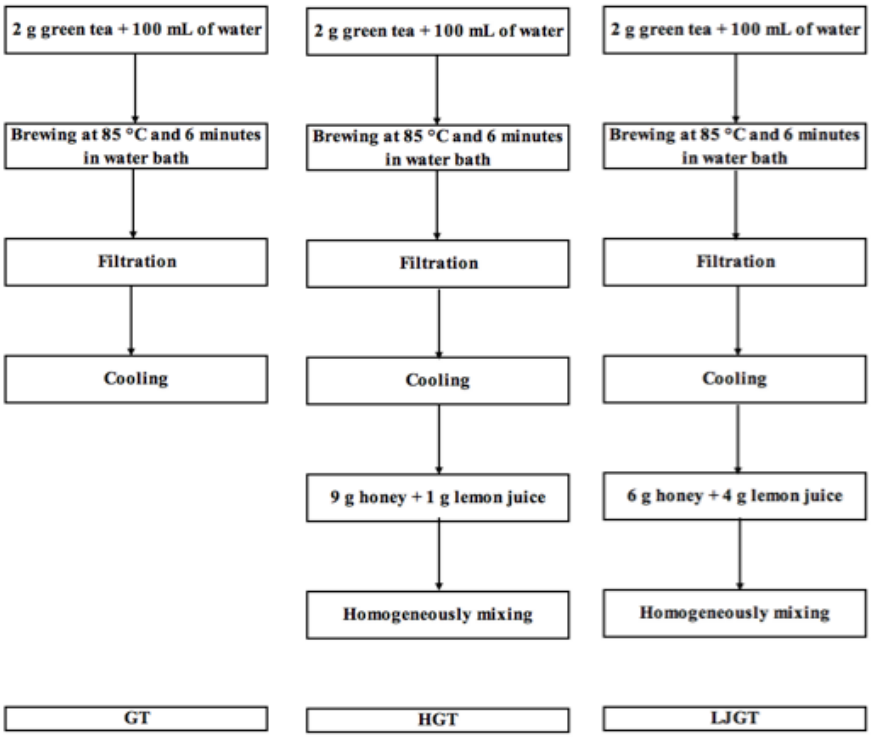

Figure 1. Production flow chart of traditionally brewed cold green teas (GT: control green tea; HGT: green tea with 9:1 ratio of honey: lemon juice; LJGT: green tea with 6:4 ratio of honey: lemon juice).

Determination of total phenolic content

The total phenolic content of the cold tea samples was determined according to the Singleton and Rossi method with some minor changes. 0.1 mL of tea sample and 0.9 mL of distilled water were added to the test tubes **[19].** The tubes were vortexed by adding 4 mL of 0.2 N Folin-Ciocalteu reagent. The samples were covered with aluminum foil and kept for 2 hours at room temperature and in a dark place. At the end of the period, the absorbances were determined by a spectrophotometer (PG Instruments, T60UV-Visible, UK) at 760 nm. The total phenolic content of the samples

was calculated using the standard curve of gallic acid. Results were expressed in mg gallic acid equivalent (GAE) per 1mL of tea sample. Analysis of total phenolic content was done in 4 parallels for each sample.

Determination of DPPH Radical-Scavenging Activity

Antioxidant activities of cold tea samples were determined according to the Brand – William et al. method with some modifications. 0.1 mL of tea samples were taken into test tubes and 3.9 mL of 6 x 10^5 DPPH was added to each tube and then vortexed **[20].** The samples were wrapped with aluminum foil and kept in a dark place for 30 minutes at room temperature. The absorbances of mixtures were measured by the spectrophotometer in the 515 nm wavelength against blank. DPPH radical scavenging activity (%) was calculated according to the following formula:

DPPH radical scavenging activity (%) = ($Absorbance_{Blank}$ – $Absorbance_{sample}$) / $Absorbance_{blank}$) x 100

Sensory analysis

In the food industry, sensory tests are applied for the acceptability of the product **[21].** The hedonic scale is the most widely used method for evaluating product acceptance among sensory tests **[22].** The use of hedonic scales allows one sample to be preferred over other samples **[21].** In this way, panelists' comments and likes about the product are evaluated **[22].** A 5-point hedonic scale (1 = Very bad, 2 = Bad, 3 = Fair, 4 = Good, 5 = Very good) was used for sensory analysis of cold tea samples. The samples were evaluated in terms of odor, color, taste, and general acceptability. The samples were coded with randomly selected 3-digit numbers and presented for evaluation by panelists. The sensory analysis was carried out in a noiseless environment with a ventilation system and illuminated with white fluorescent light. It is important to provide appropriate environmental conditions in sensory analysis. The panelists were 17 people from Istanbul Aydın University faculty and students.

Statistical analysis

Analyzes were performed in 3 repetitions in parallel. Results are given as mean ± standard deviation. The results were evaluated statistically by

applying ANOVA (analysis of variance) and Duncan Multiple Comparison test and SPSS (IBM, SPSS Statistics, Version 19) (p=0.05).

RESULTS AND DISCUSSION

Color and pH value of tea samples

The color and pH values of the tea samples are shown in Table 1. For the consumer, color is an important criterion. As the ratio of lemon juice increased, the *L** value of samples also increased, and it was statistically significant ($p<0.05$). After adding honey and lemon juice to green tea, a decrease in *b** value and an increase in *a** value was observed, and it was statistically significant ($p<0.05$).

Table 1. Color and pH value results of tea samples.

Sampl[e]*	pH	*L**	*a**	*b**
GT	$5.68^{a} \pm 0.01$	$89.66^{c} \pm 0.67$	$-7.69^{c} \pm 0.50$	$30.52^{a} \pm 0.68$
HGT	$3.90^{c} \pm 0.01$	$96.54^{a} \pm 0.64$	$-3.38^{a} \pm 0.55$	$15.09^{cd} \pm 0.71$
LJGT	$3.15^{f} \pm 0.01$	$96.58^{a} \pm 0.58$	$-3.05^{a} \pm 0.48$	$13.97^{d} \pm 0.55$
U-GT	$5.51^{b} \pm 0.00$	$81.02^{e} \pm 0.05$	$-5.51^{b} \pm 0.01$	$19.01^{b} \pm 0.01$
U-HGT	$3.54^{d} \pm 0.01$	$87.97^{d} \pm 0.59$	$-2.90^{a} \pm 0.49$	$15.50^{c} \pm 0.63$
U-LJGT	$3.18^{e} \pm 0.00$	$91.00^{b} \pm 0.66$	$-2.81^{a} \pm 0.45$	$14.54^{cd} \pm 0.69$

***** Data with the same superscript in the same column does not show a significant difference according to Duncan's test ($p<0.05$).

It was found that green tea samples (GT, U-GT) had the highest, honey-dense samples had the medium (HGT, U-HGT) and lemon juice-dense samples had the lowest (LJGT, U-LJGT) pH level and it was found statistically significant ($p<0.05$). It is believed that the reason for this change is the added products and the proportions of these products. It is known that the pH of honey is low (3.2-4.5) **[23].** For this reason, the sample containing a high amount of honey is more acidic than the green tea sample. It is known that the pH of lemon juice is 2.9 on average **[24].** Therefore, it is explained that the sample where the lemon juice is dense has a lower pH than the sample where the honey is dense.

Total dry matter results of tea samples

The total dry matter results of tea products are shown in Table 2. The results of each product revealed statistically significant differences ($p<0.05$). For all samples, it was observed that the traditional method had a higher dry matter ratio than the ultrasound method. Ultrasound extraction time can be extended to achieve the level of solids transition in the conventional method. Among the product types, it has been proven that HGT has the highest dry matter and GT has the lowest dry matter. Because honey is a low-moisture food with an average moisture content of 17% **[23].** The reason why LJGT contains higher dry matter than GT can be because lemon contains 8.4% dry matter **[15].**

Table 2. Total dry matter results of tea samples.

Sample	Total Dry Matter* (%)
GT	$0.53^{e} \pm 0.11$
HGT	$4.82^{a} \pm 0.08$
LJGT	$3.62^{c} \pm 0.08$
U-GT	$0.32^{f} \pm 0.004$
U-HGT	$4.60^{b} \pm 0.13$
U-LJGT	$2.94^{d} \pm 0.06$

* Data with the same superscript in the same column does not show a significant difference according to Duncan's test ($p<0.05$).

Phenolic material and antioxidant activity

The total phenolic substance and antioxidant activity results of tea samples are shown in Table 3. Although there is no statistically significant difference between the U-HGT and U-LJGT samples, the other samples showed a statistically significant difference ($p<0.05$). The most effective result was found in the LJGT sample. The ultrasonic extraction method did not show a positive effect in terms of extraction of phenolic compounds compared to the traditional brewing method.

Honey contains lots of bioactive constituents, such as enzymes (glucose oxidase, diastase, invertase, catalase, and peroxidase), ascorbic acid, vitamins, proteins, and organic acids **[25].** It is known that especially vita-

min C and glucose oxidase have antioxidant activity in honey **[26].** In our study, there was no increase in the total phenolic content of the honey-added samples, while it was observed that the antioxidant activities increased. It can be concluded that not all antioxidant activities in honey come from the phenolic property. This information explains the relation between the antioxidant level and total phenolic compound.

Table 3. Phenolic substance and antioxidant activity results of tea samples.

Sample	Total Phenolic Substance* (mg GAE/ 1mL tea)	Antioxidant Activity* (DPPH radical scavenging activity, %)
GT	$285.76^{b} \pm 2.80$	$62.42^{b} \pm 6.15$
HGT	$277.65^{c} \pm 1.64$	$89.03^{a} \pm 2.38$
LJGT	$300.76^{a} \pm 3.34$	$91.46^{a} \pm 1.94$
U-GT	$247.48^{d} \pm 2.03$	$53.80^{c} \pm 3.58$
U-HGT	$237.37^{e} \pm 4.10$	$90.61^{a} \pm 0.56$
U-LJGT	$237.32^{e} \pm 2.69$	$92.72^{a} \pm 0.57$

*Data with the same superscript in the same column does not show a significant difference according to Duncan's test ($p<0.05$).

When the antioxidant activity results of the samples were examined, it was determined that the U-LJGT had the highest value. However, it is seen that there is no statistically significant difference between LJGT and U-HGT ($p>0.05$). The lowest value belongs to the U-GT and then the GT sample. The lowest value reached in green tea samples shows the effect of added ingredients.

Sensory analysis results of tea samples

Sensory evaluation results of tea samples are given in Table 4 and Figure 2. There is no statistically significant difference ($p>0.05$) in terms of odor and color for all tea samples. A significant difference in taste was found in the samples with honey and lemon juice compared to the green tea sample. When evaluated in terms of general aspect, there is a statistically significant difference in the example of the high amount of lemon juice compared to the control green tea sample. The addition of honey and lemon to green

tea has been shown to improve sensory properties.

Table 4. Sensory analysis results of tea samples.

Sample	Odor	Color	Taste	General Aspect
GT	$4.12^{a} \pm 0.86$	$4.35^{a} \pm 0.79$	$3.12^{b} \pm 0.93$	$3.59^{b} \pm 0.71$
HGT	$4.06^{a} \pm 0.97$	$4.12^{a} \pm 0.93$	$3.88^{a} \pm 0.86$	$4.12^{ab} \pm 0.70$
LJGT	$4.29^{a} \pm 0,99$	$3.94^{a} \pm 0.83$	$3.88^{a} \pm 0.93$	$4.18^{a} \pm 0.88$

* Data with the same superscript in the same column does not show a significant difference according to Duncan's test ($p<0.05$).

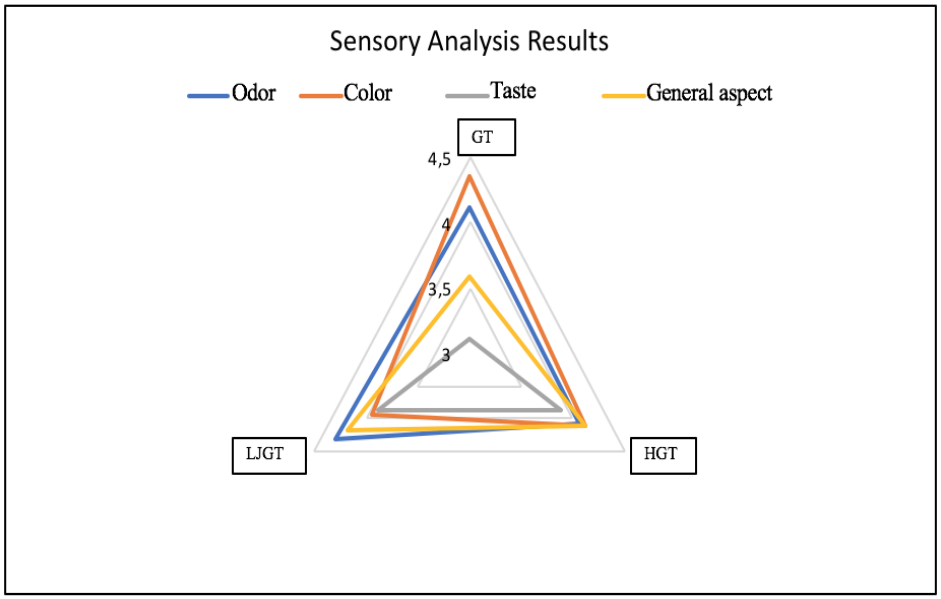

Figure 2. Sensory analysis results diagram.

CONCLUSION

Cold tea is one of the drinks that is widely consumed in Türkiye and around the world. Various additives such as colorants, sweeteners, synthetic antioxidants, and preservatives are used in the products on the market. The interest in the consumption of healthy food products is increasing daily.

It is proven that the tea formulations prepared in this study are promising for the industry. When the total phenolic substance, antioxidant activity, and sensory analysis results were evaluated, it was concluded that the best formulation was green tea containing a high amount of lemon juice. Therefore, adding honey, lemon peel, and lemon juice to green tea, can meet the market needs because of the high nutritional and good sensory properties.

REFERENCES

[1] Üstün, Ç., & Demirci, N. (2013). Çay bitkisinin (*Camellia sınensıs* L.) tarihsel gelişimi ve tıbbi açıdan değerlendirilmesi. Mersin Üniversitesi Tıp Fakültesi Lokman Hekim Tıp Tarihi ve Folklorik Tıp Dergisi, 3(3), 5-12.

[2] Owuor, P.O. (2014). Tea (Eds. Bamforth, C.W. & Ward, R.E). In The Oxford Handbook of Food Fermentations. Oxford University Press, NY, USA.

[3] Wang, C. C., & Lee, T. E. (2015). Optimization design and application of composite ultrasonic extraction method for effective constituents of green tea. Advances in Mechanical Engineering, 8(1), 1687814015624847.

[4] Alam, M., Ali, S., Ashraf, G. M., Bilgrami, A. L., Yadav, D. K., & Hassan, M. I. (2022). Epigallocatechin 3-gallate: From green tea to cancer therapeutics. Food Chemistry, 132135.

[5] Raygaert, W. (2017). An update on the health benefits of green tea. Beverages, 3, 1-14.

[6] Burucu, V., & Bal, H. S. G. (2017). Türkiye'de arıcılığın mevcut durumu ve bal üretim öngörüsü. Tarım ekonomisi araştırmaları dergisi, 3(1), 28-37.

[7] Fakhlaei, R., Selamat, J., Khatib, A., Razis, A. F. A., Sukor, R., Ahmad, S., & Babadi, A. A. (2020). The toxic impact of honey adulteration: A review. Foods, 9(11), 1538.

[8] Meo, S. A., Al-Asiri, S. A., Mahesar, A. L., & Ansari, M. J. (2017). Role of honey in modern medicine. Saudi journal of biological sciences, 24(5), 975-978.

[9] Alvarez-Suarez, J. M., Gasparrini, M., Forbes-Hernández, T. Y., Mazzoni, L., & Giampieri, F. (2014). The composition and biological activity of honey: a focus on Manuka honey. Foods, 3(3), 420-432.

[10] Bogdanov, S. (2012). Honey as nutrient and functional food. Proteins, 1100, 1400-2700.

[11] Ball, D. W. (2007). The chemical composition of honey. Journal of chemical education, 84(10), 1643.

[12] Bogdanov, S., Jurendic, T., Sieber, R., & Gallmann, P. (2008). Honey for nutrition and health: a review. Journal of the American college of Nutrition, 27(6), 677-689.]

[13] Subramanian, R., Umesh Hebbar, H., & Rastogi, N. K. (2007). Processing of honey: a review. International Journal of Food Properties, 10(1), 127-143.

[14] Mohanapriya, M., Ramaswamy, L., & Rajendran, R. (2013). Health and medicinal properties of lemon (Citrus limonum). International Journal of Ayurvedic and Herbal Medicine, 3(1), 1095-100.

[15] Jiang, H., Zhang, W., Xu, Y., Chen, L., Cao, J., & Jiang, W. (2022). An advance on nutritional profile, phytochemical profile, nutraceutical properties, and potential industrial applications of lemon peels: A comprehensive review. Trends in Food Science & Technology.

[16] Ateş, E. (2017). Ultrason Destekli Ekstraksiyon Uygulamasının Yeşil Çay Ekstraktının Fizikokimyasal ve Fitokimyasal Özellikleri Üzerine Etkisi. (Master's thesis, Fen Bilimleri Enstitüsü).

[17] Jin, Y., Zhao, J., Kim, E. M., Kim, K. H., Kang, S., Lee, H., & Lee, J. (2019). Comprehensive investigation of the effects of brewing conditions in sample preparation of green tea infusions. Molecules, 24(9), 1735.

[18] Yeşilören Akal, G. (2019). Siyah havuç posasından antosiyanin ekstraksiyonu. (Master's thesis, Ankara Üniversitesi).

[19] Singleton, V. L., & Rossi, J. A. (1965). Colorimetry of total phenolics with phosphomolybdic-phosphotungstic acid reagents. American journal of Enology and Viticulture, 16(3), 144-158.

[20] Brand-Williams, W., Cuvelier, M. E., & Berset, C. L. W. T. (1995). Use of a free radical method to evaluate antioxidant activity. LWT-Food science and Technology, 28(1), 25-30.

[21] Bergara-Almeida, S., Aparecida, M., & Da Silva, A. P. (2002). Hedonic scale with reference: Performance in obtaining predictive models. Food Quality and Preference, 13(1), 57-64.

[22] Barış, O. (2017). Gıdalarda ürün ve marka tercihinde tüketici algısının belirlenmesi: Bursa ili örneği (Master's thesis, Uludağ Üniversitesi).

[23] da Silva, P. M., Gauche, C., Gonzaga, L. V., Costa, A. C. O., & Fett, R. (2016). Honey: Chemical composition, stability and authenticity. Food chemistry, 196, 309-323.

[24] Asefi, N., & Artık, N. Limon Suyu ve Limon Konsantresi Üretimi ve Bileşim Unsurları Üzerine Araştırma. Gıda, 17(5).

[25] Erejuwa, O. O., Sulaiman, S. A., & Ab Wahab, M. S. (2012). Honey: a novel antioxidant. Molecules, 17(4), 4400-4423.

[26] Khalil, M. I., Sulaiman, S. A., & Boukraa, L. (2010). Antioxidant properties of honey and its role in preventing health disorder. The Open Nutraceuticals Journal, 3(1).

PRODUCTION OF PROBIOTIC JUICE IN FRUIT MATRIX

Tuğçe CEYHAN[1], İzem SÜZGÜN[2]*

Istanbul Aydın University, Faculty of Engineering,
Department of Food Engineering
**Corresponding author: izemsuzgun@stu.aydin.edu.tr*
ORCID number: [1]0000-0002-7189-7439, [2]0000-0002-7610-8996

ABSTRACT

Probiotics are organisms that contribute to the intestinal and digestive systems. Microorganisms that are considered commercial probiotics, most often belong to the genus *Lactobacillus*. Examples of probiotic *Lactobacillus* species are *Lactobacillus acidophilus*, *L. rhamnosus, L. casei, L. plantarum, L. bulgaricus, L. Delbrueckii,* and *L. helveticus*. *Lactobacillus* species are generally recognized as safe (GRAS) organisms. Lactic acid bacteria (LAB) are widely used in the production of fermented foods and beverages because they produce the desired properties in food and beverages thanks to their fermentation properties. They play an important role in ensuring food safety and extending the shelf life of food by producing inhibitory metabolites against other undesirable microorganisms while developing in food. In recent years, non-dairy products, and carriers of probiotics have become more and more used. The main reasons for this are lactose intolerance, vegetarianism, and the fact that people want to move away from animal foods. Juices are non-dairy matrices for probiotics and are compatible with popular dietary options.

Keywords: *Probiotic, Fruit juice, Lactic acid bacteria (LAB).*

INTRODUCTION

The intestinal microbiota in humans plays a specific role in the physiology and metabolism of the host **[1]**. Probiotics, on the other hand, are living microorganisms that have a positive effect on human health by being present in the digestive tract of the body. Probiotics are defined as; "live microorganisms which when administered in adequate amounts confer a

[1]Geliş Tarihi: 11.05.2021 Kabul Tarihi: 07.06.2021
DOI: 10.17932/IAU.IJFER.2015.003/ijfer_v07i2002

health benefit on the host." **[2]**. But in addition to the fact that probiotics are consumed regularly and their number is sufficient, these bacteria can be found in difficult environmental conditions in the digestive tract (gastric acidity, bile salts, enzymes, etc.) they must be able to maintain their viability and reach and colonize the appropriate number of intestines **[3]**.

Probiotics have usually been used in fermented dairy products such as kefir, yoğurt, and so on. But for many reasons, such as lactose intolerance, and vegetarianism, which have appeared in recent years, there is an increasing interest in non-dairy probiotic products. Being the main source of water-soluble vitamins, minerals, dietary fibers, and phytochemicals, fruits play an important role in the human diet **[4].** Juices are useful food products for health and are regularly consumed by many people. Due to the rich content of juices, the absence of allergenic effects, and the fact that they can be consumed by everyone, they are quite suitable for the use of probiotic cultures **[5]**. In addition, the fruits have the appropriate matrix for probiotic adhesion. Because of this, it is thought that fruits with high fiber content, such as apples, will have a protective effect on probiotic microorganisms **[6]**. In accordance with this information; various studies have been conducted on the survival of probiotic LAB in fruit juices such as apple juice **[7],** grape juice **[8, 9],** pomegranate juice **[10],** as well as in juices. In different studies, it has been stated that fruits and vegetables are a good food matrix for probiotic bacteria **[11, 12].**

Probiotics in Fruit Matrix

The main purpose of probiotic foods that have positive effects on health is for microorganisms to stay in the body longer by being taken into the body with different matrices and kept in the intestinal microflora **[13].** Most of the probiotic microorganisms belong to the genera *Lactobacillus, Enterococcus, and Bifidobacterium*, which are lactic acid bacteria (LAB). Some species of *Saccharomyces, Streptococcus, Enterecossuc, Pediococcus, Lactococcus, Leunocostoc, Bacillus,* and *Clostridium* can also be used as yeast and other species of bacteria **[14-18].** LAB are a group of microorganisms that produce lactic acid by fermenting Gram-positive, immobile, non-spore-forming, rod-shaped or coke-shaped, catalase-negative, aerotolerant, acid-resistant, carbohydrates, and high alcohols **[19]**. The largest group in the LAB is the genus *Lactobacillus* **[20, 21].** As for the species that are widely studied probiotics, they usually belong to the genus *Lacto-*

bacillus. Probiotic strains belonging to the genus *Lactobacillus*, which are naturally found in the human gastrointestinal tract, are considered GRAS (generally recognized as safe) **[22]**.

The most important factors affecting the number of living cells contained in probiotics are the fermentation time of the product, the storage temperature, and the oxygen content **[23]**. A microorganism that will be used as a probiotic must remain alive during its passage through the digestive tract. For this reason, it must be resistant to enzymes such as lysozyme, pepsin, and pancreatin and survive in the gastric environment of the stomach (pH 1.5-3.0). Probiotic microorganisms, thanks to their resistance to enzymes, can reach the intestine without being adversely affected by gastric acidity **[24]**. Compared to dairy products, the survival of probiotics in the fruit matrix is more complicated. Because of the acidic structure of the fruits, LAB needs to protect themselves from the high acidity in this environment **[25]**. Bile salts are synthesized from cholesterol in the liver and secreted from the gallbladder into the duodenum (500-700 ml/day) **[26].** These acids then undergo chemical modifications (dehydrogenation, deconjugation, deglucuronidation, and dehydroxylation) due to microbial activity in the colon **[27-29].** All conjugated and unconjugated bile salts have an antibacterial effect for strains of *Escherichia coli*, *Klebsiella* and *Enterococcus* species **[26, 30, 31].** Unconjugated bile salts have higher antimicrobial activity. The sensitivity of Gram-positive bacteria to bile salts is higher than that of Gram-negative bacteria **[32, 33].** In vitro resistance tests have shown that bifidobacteria are more resistant to bile salts than lactobacilli **[34, 35].**

By adding prebiotic components to probiotic juices, improvement in sensory and physicochemical properties and an increase in probiotic survival occur **[5, 36].** But it is impossible not to mention the definite positive effect of the addition of prebiotics on probiotic survival. Because while some studies **[7]** support the positive effect of prebiotics on the development of probiotics, some studies **[5, 38, 39]** have stated that there is no positive effect**.** In addition, it has been stated that processing techniques such as peeling and small-piece separation applied to fruits and vegetables will have a positive effect on probiotic beverage production by releasing cellular content in terms of minerals, sugars, vitamins, and other nutrients and therefore creating a richer substrate environment for probiotics **[40, 41]**. Table 1 shows the probiotic juice studies produced using different fruit matrices and microorganisms.

Table 1. Probiotic application in fruit juices

Plant matrix	Microorganisms	References
Apple	*Lactobacillus casei*	**[42]**
Apple	*L. paracasei*	**[7]**
Cashew apple	*L. casei*, *Bifidobacterium breve*, *B. infantis*	**[43]**
Grape	*L. paracasei*	**[44]**
White grape	*L. paracasei*, *L. casei*-01	**[9]**
Grape	*L. plantarum* PTCC, *L. delbruecki* PTCC 1737i, *L. rhamnosus* PTCC 1657	**[45]**
Citrus	*L. pentosus* MU-1, *L. plantarum* SI-1	**[46]**
Orange	*Pediococcus acidilactici*, *L. monocytogenes*	**[47]**
Orange	*L. rhamnosus*	**[48]**
Orange	*L. paracasei*	**[5]**
Orange	*L. case*i-01	**[49]**
Pineapple	*P. pentosaceus* LaG1, *L. rhamnosus* GG, *P. pentosaceus* LBF2	**[50]**
Pineapple	*Lactobacillus* and *Bifidobacterium* spp.	**[51]**
Mango	*L. acidophilus*	**[52]**
Mango	*L. plantarum, L. casei, L. acidophilus, L. delbrueckii*	**[53]**
Mango	*L. bulgaricus* S1, L. *plantarum* Lp-115, *Streptococus thermophilus* 6063	**[54]**
Pomegranate	*L. plantarum,* , *L. paracasei, L. delbruekii, L. acidophilus*	**[55]**
Pomegranate	*L. plantarum*	**[56]**
Pomegranate	*L. plantarum* PTCC 1745, *L. delbruecki*i: PTCC 1333	**[10]**
Watermelon	*L. helveticus, L. acidophilus*, *L. plantarum*	**[57]**
Melon	*L. casei* B-442	**[58]**
Cornelian cherry	*L. plantarum* ATCC 14917	**[59]**

Cornelian cherry	*L. casei* T4	**[13]**
Blueberry and blackberry	*L. plantarum, Streptococcus thermophilus*, *B. bifidum*	**[60]**
Blueberry	*L. plantarum* J26	**[61]**
Prickly pears	*L. fermentum*	**[62]**
Peach	*L. plantarum*, *L. delbrueckii*, *L. casei*	**[63]**
Noni	*L. casei, L. plantarum*, *B. longum*	**[64]**
Carrot, beet and apple	*L. casei*	**[65]**
Cupuassu	*L. casei*	**[66]**

In the study of Pereira et al. **[42],** optimizing the living standards of *L. casei* NRRL B-442 in cashew apple juice, determining the appropriate amount of inoculum and the fermentation time were investigated. As a result of the analysis, they observed that there was no loss of vitality of *L. casei* in cold storage for 42 days in apple juice and that the color, which is one of the most important factors after flavor, was preserved in the juice. For this reason, it has been reported that cashew apple juice fermented with *L. casei* is a healthy alternative to functional foods containing probiotics.

In the study of Pimentel et al. **[7],** the effects of using oligofructose or sucralose instead of sugar on probiotic apple juice were investigated by adding *L. paracasei* ssp. Apple juice containing oligofructose showed less sweetness than that containing sucrose. It has been observed that sucralose apple juices have a light color compared to sucrose apple juice. The addition of probiotics has increased the turbidity of apple juice. The increase in its turbidity did not affect the general acceptability of apple juice. It has been reported that with *L. paracasei*, synbiotic apple juice can be created that creates a sensory profile (excluding turbidity and particle presence) and acceptance level close to the sucrose-added apple juice.

In the study of Leite et al. **[43],** cashew apple juice containing gluco-oligosaccharides, dextran, and tagatose has been confirmed to exhibit prebiotic properties after in vitro digestion. These components have reached the digestive tract and are used as a source of carbon by probiotic bacteria (*L. casei* NRRL B-442, *B. breve* NRRL B-41408, and *B. infantis* NRRL B-41661). For all the bacteria studied, a different growth was observed

against substrates with prebiotic properties (oligosaccharides, dextran, and tagatose). It has been reported that the substrates used support probiotic microbial growth.

In the study of Silva & Ferrari **[44]**, the effect of inulin on the viability of microorganisms was observed in grape juice produced using *L. paracasei.* In order to observe the effect of inulin, 3 formulations were compared: grape juice without probiotics and inulin, grape juice with *L. paracasei* (7.5%), and grape juice with *L. paracasei* (7.5%, v/v) / inulin (10%, w/v), which did not participate in inulin, and grape juice with *L. paracasei* (7.5%, v/v). The probiotic grape juices produced were stored at 4ºC for 28 days and subjected to physical and microbiological analysis. Although the pH and total acidity values of dec grape juices ranged between 3.28-3.31 and 0.193-0.209 g/ml, respectively, no statistically significant difference was observed. It has been determined that the amount of soluble solids in the juice of probiotics containing inulin is greater. Grape juice has proved to be a promising plant-based matrix for the production of a probiotic drink, as it is able to ensure the viability of *L. paracasei* above 6 log CFU/ml by the end of storage. The results of the study showed that *L. paracasei* is able to survive in grape juice even without the addition of a prebiotic component.

In the study of Okina et al. **[9],** probiotic white grape juice was made using lyophilized *L. paracasei* culture (2% v/v), and the culture was compared with non-culture-added juice. It has been determined that the white grape juice produced by the added probiotic culture has a darker color and a lower total phenolic amount. As a result, it was emphasized that probiotic white grape juice prepared using *L. paracasei* is an alternative for individuals who do not consume/can not consume dairy products and can resist the probiotic effect (21 days at 4°C) and gastrointestinal conditions (28 days at 4°C). The study also stated that white grape juice is a good carrier of probiotics for *L. paracasei*.

In the study of Manganji vd. **[45],** by adding *L. delbrueckii, L. Plantarum,* and *L. rhamnosus* to pasteurized grape juice, its change in microbiological and sensory properties was studied for 4 weeks at 4°C. According to the study, the resistance of *L. rhamnosus* and *L. delbrueckii* to refrigerator conditions was found to be better compared to *L. plantarum*. A decrease in probiotic levels was observed starting from the 1st to the 28th day, but

the least decrease was observed in *L. rhamnosus*. In addition, the sensory properties of grape juice added to L rhamnosus and kept in refrigerator conditions for 4 weeks were found to be at a higher level of acceptability compared to others. According to the findings obtained as a result of the study, it is recommended to produce probiotic grape juice with the addition of *L. rhamnosus*, whose vitality is higher and organoleptic properties are more desired.

In the study of Yuasa et al. **[46],** *L. plantarum* SI-1 and *L. pentosus* MU-1 were inoculated into three different citrus juices and fermented. It has been reported that there is a decrease in the number of living bacteria in citrus juices, but their taste and other sensory properties have not changed.

In the study of de Oliveira Vieira et al. **[47],** it has been confirmed that oranges can be used as an ideal food matrix for a probiotic drink. It has been reported that *P. acidilactici* CE51 is suitable for the production of probiotic orange juice. It has also been reported that *P. acidilactici* can be used to provide control against *L. monocytogenes*.

In the study of Sengun et al. **[48],** it has been reported that orange-based juices are suitable substrates for the development of *L. rhamnosus*, and these products can be considered probiotic products. In the same study, they reported that the addition of stinging nettle (*Urtica dioica* L.) together with *L. rhamnosus* did not significantly increase the viability of *L. rhamnosus* in cold-stored orange juice, but increased the total content of phenolic substances and slowed the rate of decline of antioxidant activity during storage. As a result of the study, it was reported that the production of probiotic orange juice using *L. rhamnosus* and stinging nettle was successfully completed.

In the study of Costa et al. **[5],** the effect of the use of oligofructose or ascorbic acid on the viability of *L. paracasei* has been investigated. Probiotic juices produced high probiotic culture viability during the 28-day cold storage period. However, oligofructose or ascorbic acid did not have a protective effect on probiotic strains. As a result, it has been reported that orange juice is a suitable substrate for the development of *L. paracasei*.

In the study of Miranda et al. **[49],** the effect of the addition of *L. casei* to orange juice on the quality of the juice was studied. The cultures used in

the study were added to orange juice directly, activated, and encapsulated. As a result of the direct addition of commercial cultures directly, it was found that the probiotic drink is more similar to pure fruit juice in terms of its physical, chemical, and sensory properties. At the same time, it was the drink that had the most suitable products in terms of volatile compounds. As a result of the addition of activated cultures, an increase in the content of organic acids (acetic, lactic, and citric) was observed, while the volatile components decreased and were less appreciated sensually. By adding encapsulated cultures, products with a high consistency index were obtained, while lower organic acid and sensory results were determined.

In the study of AdebayoTayo & Akpeji **[50],** probiotic LAB (*P. pentosaceus* LaG1, *L. rhamnosus* GG, and *P. pentosaceus* LBF2) of fermented pineapple juice produced by using single and multiple the viability of LAB, lactic acid production, vitamin C has been reported to increase the growth and antagonistic potential. It has been reported that the probiotic fermented pineapple juice produced in this way can be a beneficial drink for human health.

In the study of Nguyen et al. **[51],** it has been reported that pineapple juice is a good substrate for the development of probiotic bacterial strains such as *Lactobacillus* and *Bifidobacterium*, however, it is more appropriate to use *L. plantarum* for probiotic pineapple juice.

In the study of Ryan et al. **[52],** the effect of mango juice and milk-based fermented beverage on the viability of probiotic strains has been investigated. In the study, *L. acidophilus* strain was detected in different concentrations of mango juice in cow's milk (formulations containing 0%, 10%, 20%, 30%, and 40% mango juice (w/w)) were added and kept at 4 °C for five weeks. It has been determined that the concentration of 10% mango juice increases the viability of probiotic cultures. Similarly, it was determined that 10% mango juice increases the probiotic tolerance of gastrointestinal digestion under artificial conditions. It has been reported that increasing the concentration of mango juice from 20% to 40% increases sensory scores and therefore is more appreciated.

In the study of Reddy et al., **[53],** it has been observed that four LAB such as *L. plantarum, L. delbrueckii, L. casei* and *L. acidophilus* can use mango juice for cell synthesis and lactic acid production without pH adjustment.

All of the LAB used in the study survived under conditions of low pH and high acidity during 4 weeks of cold storage at 4 °C. It has been reported that mango juice can be used as one of the non-dairy raw materials for LAB, and the product can be used as a probiotic drink for vegetarians.

In the study of Wang et al. **[54],** ultraviolet-assisted ultrasonic pretreatment for probiotic mango juice containing *L. bulgaricus* S1, 6063 *S. Thermophilus,* and *L. plantarum* Lp-115 strains have been applied as pre-processing. It has been reported that probiotic mango juices produced using probiotic strains have positive effects such as better storage stability and longer shelf life compared to probiotic mango juice produced without pretreatment.

In the study of Mousavi et al. **[55]**, the suitability of pomegranate juice produced by probiotic LAB for use as a probiotic beverage has been studied. Pomegranate juice was fermented at 30°C for 72 hours. *L. delbruekii* and *L. plantarum* were able to survive the first 2 weeks of storage at 4°C, while *L. acidophilus* and *L. paracasei* were observed to lose their viability after the second week in the same conditions. It has been reported that fermented pomegranate juice is a suitable medium for probiotic cultures to increase.

In the study of Mantzourani et al. **[56]**, It was tried to obtain a low-alcohol functional pomegranate juice drink by adding *L. plantarum*. Fermented pomegranate juice has had a high total phenolic content and antioxidant activity during the storage period compared to pomegranate juice which has not undergone a fermentation process. *L. plantarum* has maintained its vitality and increased in fermented pomegranate juice. As a result of these studies, it was reported that *L. plantarum* can be used for the development and production of a new functional drink.

In the study of Oruç & Çakır **[57]**, for the fermentation of watermelon juice, *L. helveticus, L. Acidophilus,* and *L. plantarum* were used. Watermelon juice was fermented at 37°C for 18 hours. During the fermentation process, total LAB count, titration acidity, pH, and water-soluble dry matter analyses of watermelon juices were performed. It was determined that the LAB numbers increased significantly in all watermelon juice samples. It has been reported that watermelon juice is a favorable environment for the development of LAB, and with fermentation, there is an increase in the values of phenolic substances and antioxidant activity of watermelon juice.

In the study of Fonteles et al. **[58],** melon juice fermented with *L. casei* was subjected to ultrasonic processing. During the 42-day storage period in fermentation without the use of preservatives and heat treatment, it was not observed that the bacterium did not lose its viability and that there was no deterioration in the juice. It has been reported that melon juice fermented with the support of ultrasonic processing can be considered a low-calorie drink, which is a potential new source of probiotic bacteria.

In the study of Mantzourani et al. **[59]**, *L. plantarum* ATCC 14917 was used in cornelian cherry juice fermentation. Cornelian cherry juice was fermented for 24 hours and kept at 4 °C for 4 weeks. As a result, it has been shown that *L. plantarum* immobilized on the wheat bran carrier used can be used in the production of a low-alcohol grade cranberry beverage. As a result of sensory evaluations, it was determined that there is no unpleasant/undesirable taste in fermented cranberry juice. It has been reported that fermented cornelian cherry juice is a suitable medium for probiotic cultures to increase.

In the study of Nematollahi et al. **[12],** cornelian cherry juice with a pH of 2.6 is harmful to probiotic cultures, and probiotic species that are resistant to even the worst conditions have been found to disappear completely after a maximum of 7 days of cold storage. The viability of *L. casei* T4, which was applied by adjusting to pH 3.5, was even greater after 28 days than at the beginning. As a result, it has been reported that *L. casei* T4 will be a strong and resistant strain for production in beverages with harsh conditions, such as fruit juice and beer.

In the study of Wu et al. **[60]**, it has been revealed that potential probiotic strains (*L. plantarum, B. Bifidum, and S. thermophilus*) have effective values on the metabolism of phenolic acids, biotransformation of organic acids, and sensory properties in blueberry and blackberry juices. It has been reported that the study conducted can help produce potential non-dairy probiotic drinks.

In the study of Zhang et al. **[61]**, blueberry juice has been fermented using *L. plantarum* J26 as the fermentative strain for the investigation of its functional properties. Functional ingredients fermented blueberry juice (cyanide, chloride, petunidin, and pelargonidin peonid), and beneficial activities (α-glucosidase and α-amylases inhibition) observed an increase in

LAB can produce a beverage with functional and also used blueberry has been reported.

In the study of Panda et al. **[62]**, total LAB count, titration acidity, pH and water-soluble dry matter analyses were performed by fermenting prickly pear juice with *L. fermentum* at 28°C for 48 hours. As a result of the study, it was confirmed that it is acceptable research to prevent prickly pears from losing their properties and the value of pear juice. It has also been reported that prickly pear juice is a new functional drink that contains antioxidants and phytochemicals that are beneficial to health.

In the study of Pakbin et al. **[63],** probiotic peach juice production was carried out by fermentation at 30 °C for 24 hours using *L. plantarum* DSMZ 20179, *L. delbrueckii* DSMZ 15996 and *L. casei* DSMZ 20011 strains. Changes in pH, titratable acidity, sugar content, and the number of living cells were studied during fermentation. As a result of the study, *L, casei* was not considered suitable for cold storage, but it was found that *L. delbrueckii* is suitable for the production of probiotic beverages. Therefore, it has been reported that peach juice can be a healthy beverage alternative for vegetarians and consumers with food allergies.

In the study of Wang et al. **[64]**, *L. casei, L. Plantarum,* and *B. longum* were used to produce probiotic noni (*Morinda citrifolia*) juice. It has been observed that *L. casei* produces less lactic acid than *B. longum* and *L. plantarum*. It has been observed that *L. plantarum* and *B. longum* survive at low pH in cold storage at 4° C for 4 weeks, and cell viability has not been observed in *L. casei* after 3 weeks. As a result of the study, it was reported that *L. plantarum* and *B. longum* are optimal probiotics for fermentation with noni juice.

In the study of Zandi et al. **[65]**, a mixture of carrot, beet, and apple juices was infused with *L. casei* to produce a fermented beverage. A mixture of carrot, beet, and apple juices and a suspension of *L. casei* was prepared and added to the mixture of juices by 20%, 30%, and 40%, respectively. In all applications, during the fermentation process, the number of probiotic bacteria increased with the use of sugars and nutrients contained in the juice, while the values of sugar and brix decreased. It has been reported that the sample with a concentration of 40% and $1.5x10^6$ CFU/ml *L. casei* has maximum cell viability during 4 weeks of storage at 4° C and is suitable

for use as a probiotic beverage.

In the study of Pereira et al. **[66],** cupuassu (tropical fruit native to the Brazilian Amazon) was fermented for 18 hours at 30 °C, pH 5.8 by adding *L. casei* to the juice. During fermentation, the antioxidant activity and the number of *L. casei* increased. After fermentation, the organic acids contained in cupuassu water increased. As a result, the components of cupuassu water (natural sugars and organic acids) were found to be suitable for the development of probiotic bacteria. It has been reported that the probiotic cupuassu drink is considered an alternative functional food.

CONCLUSION

Probiotics are organisms that contribute to the intestinal and digestive systems. Probiotics, which are usually used in yogurt, kefir, and similar fermented products, have also started to be used in fruits and vegetables that are considered to be the different probiotic matrices. In different studies, it has been shown that many fruits are a suitable environment for probiotics. The rich content of fruits, the absence of allergenic effects, and the fact that they can be consumed by everyone are quite suitable for the use of probiotic cultures. It will be useful to investigate the production of probiotic functional juice in different types of fruits and with different microorganisms.

REFERENCES

[1] Scholtens, P. A., Oozeer, R., Martin, R., Amor, K. B., & Knol, J. (2012). The early settlers: intestinal microbiology in early life. Annual Review of Food Science and Technology, 3, 425-447.

[2] FAO/WHO (2006). Probiotics in Food: Health and Nutritional Properties and Guidelines for Evaluation. FAO Food Nutrition Pap. 85. Rome: World Health Organization and Food and Agriculture Organization of the United Nations.

[3] İnanç, N., Şahin, H., & Çiçek, B. (2005). Probiyotik ve prebiyotiklerin sağlık üzerine etkileri. Erciyes Tıp Dergisi, 27(3), 122-127.

[4] Gebbers, J. O. (2007). Atherosclerosis, cholesterol, nutrition, and statins–a critical review. GMS German Medical Science, 5.

[5] da Costa, G. M., de Carvalho Silva, J. V., Mingotti, J. D., Barão, C. E., Klososki, S. J., & Pimentel, T. C. (2017). Effect of ascorbic acid or oligofructose supplementation on L. paracasei viability, physicochemical characteristics and acceptance of probiotic orange juice. LWT- Food Science and Technology, 75, 195-201.

[6] Kourkoutas, Y., Xolias, V., Kallis, M., Bezirtzoglou, E., & Kanellaki, M. (2005). *Lactobacillus casei* cell immobilization on fruit pieces for probiotic additive, fermented milk and lactic acid production. Process Biochemistry, 40(1), 411-416.

[7] Pimentel, T. C., Madrona, G. S., Garcia, S., & Prudencio, S. H. (2015). Probiotic viability, physicochemical characteristics and acceptability during refrigerated storage of clarified apple juice supplemented with *Lactobacillus paracasei* ssp. *paracasei* and oligofructose in different package type. LWT-Food Science and Technology, 63(1), 415-422.

[8] Malganji, S., Sohrabvandi, S., Jahadi, M., Nematollahi, A., & Sarmadi, B. (2016). Effect of refrigerated storage on sensory properties and viability of probiotic in grape drink. Applied Food Biotechnology, 3(1), 59-62.

[9] Okina, V. S., Porto, M. R. A., Pimentel, T. C., & Prudencio, S. H. (2018). White grape juice added with *Lactobacillus paracasei* ssp. probiotic culture. Nutrition & Food Science, 48(4), 631-641.

[10] Dogahe, M., Khosravi-Darani, K., Tofighi, A., Dadgar, M., & Mortazavian, A. (2015). Effect of Process Variables on Survival of Bacteria in Probiotics Enriched Pomegranate Juice. British Biotechnology Journal, 5(1), 37-50.

[11] Nagpal, R., Kumar, A., & Kumar, M. (2012). Fortification and fermentation of fruit juices with probiotic lactobacilli. Annals of Microbiology, 62(4), 1573-1578.

[12] Nematollahi, A., Sohrabvandi, S., Mortazavian, A. M., & Jazaeri, S. (2016). Viability of probiotic bacteria and some chemical and sensory characteristics in cornelian cherry juice during cold storage. Electronic Journal of Biotechnology, 21, 49-53.

[13] Heperkan, Z. D. (2021). Gıda ve probiyotikler. In: Tıp ve Mühendislik Bakış Açısıyla Probiyotikler ve Prebiyotikler (Eds: Z.D. Heperkan & Z.Ç Kayacan), İstanbul Aydın Üniversitesi Yayınları, 71-89.

[14] Klein, G., Pack, A., Bonaparte, C., & Reuter, G. (1998). Taxonomy and physiology of probiotic lactic acid bacteria. International Journal of Food Microbiology, 41(2), 103-125.

[15] Elmer, G. W., Martin, S. W., Horner, K. L., Mcfarland, L. V., & Levy, R. H. (1999). Survival of *Saccharomyces boulardii* in the rat gastrointestinal tract and effects of dietary fiber. Microbial Ecology in Health and Disease, 11(1), 29-34.

[16] Senesi, S., Celandroni, F., Tavanti, A., & Ghelardi, E. (2001). Molecular characterization and identification of *Bacillus clausii* strains marketed for use in oral bacteriotherapy. Applied and Environmental Microbiology, 67(2), 834-839.

[17] Takahashi, M., Taguchi, H., Yamaguchi, H., Osaki, T., Komatsu, A., & Kamiya, S. (2004). The effect of probiotic treatment with *Clostridium butyricum* on enterohemorrhagic *Escherichia coli* O157: H7 infection in mice. FEMS Immunology & Medical Microbiology, 41(3), 219-226.

[18] Lebaka, V. R., Wee, Y. J., Narala, V. R., & Joshi, V. K. (2018). Development of new probiotic foods—a case study on probiotic juices. Therapeutic, Probiotic, and Unconventional Foods, 55-78.

[19] Vuyst, L.D., & Leroy, F. (2007). Bacteriocins from lactic acid bacteria: Production, purification and food applications, Journal of Molecular Microbiology Biotechnology 13, 194-199.

[20] Stiles, M. E., & Holzapfel, W. H. (1997). Lactic acid bacteria of foods and their current taxonomy. International Journal of Food Microbiology, 36(1), 1-29.

[21] Tannock, G. W. (2004). A special fondness for lactobacilli. Applied and Environmental Microbiology, 70(6), 3189–3194.

[22] Amiri, S., Rezaei Mokarram, R., Sowti Khiabani, M., Rezazadeh Bari, M., & Alizadeh Khaledabad, M. (2019). Exopolysaccharides production by *Lactobacillus acidophilus* LA5 and *Bifidobacterium animalis* subsp. *lactis* BB12: Optimization of fermentation variables and characterization of structure and bioactivities. In International Journal of Biological Macromolecules, 123, 752-765).

[23] Yoon, K. Y., Woodams, E. E., & Hang, Y. D. (2005). Fermentation of beet juice by beneficial lactic acid bacteria. LWT-Food Science and Technology, 38(1), 73-75.

[24] Shah, N.P. (2001) Functional Foods, Probiotics and Prebiotics. Food Technology, 55, 46-53.

[25] Shah, N. P. (2007). Functional cultures and health benefits. International Dairy Journal. 17(11), 1262-1277.

[26] Hofmann, A. F., Molino, G., Milanese, M., & Belforte, G. (1983). Description and simulation of a physiological pharmacokinetic model for the metabolism and enterohepatic circulation of bile acids in man. Cholic acid in healthy man. The Journal of Clinical Investigation, 71(4), 1003-1022.

[27] Hill, M.J. and Draser, B.S. (1968). Degradation of bile salts by human intestinal bacteria. Gut (9), 22-27.

[28] Shimada, K., Bricknell, K. S., & Finegold, S. M. (1969). Deconjugation of bile acids by intestinal bacteria: review of literature and additional studies. The Journal of Infectious Diseases, 273-281.

[29] Hylemon, P.B. and Glass, T.L. (1983). Biotransformation of bile acids and cholesterol by the intestinal microflora. In: Hentes D. J. (Ed) Human Intestinal Microflora in Health and Disease, 189-213. Academic Press, New York.

[30] Lewis, R., & Gorbach, S. (1972). Modification of bile acids by intestinal bacteria. Archives of Internal Medicine, 130(4), 545-549.

[31] Lee, Y. K., & Salminen, S. (1995). The coming of age of probiotics. Trends in Food Science & Technology, 6(7), 241-245.

[32] Floch, M. H., Binder, H. J., Filburn, B., & Gershengoren, W. (1972). The effect of bile acids on intestinal microflora. The American Journal of Clinical Nutrition, 25(12), 1418-1426.

[33] Tahri, K., Grille, J. P., & Schneider, F. (1996). *Bifidobacteria* strain behavior toward cholesterol: coprecipitation with bile salts and assimilation. Current Microbiology, 33(3), 187-193.

[34] Dunne, C. (2001). Adaptation of bacteria to the intestinal niche: pro-

biotics and gut disorder. Inflammatory Bowel Diseases, 7(2), 136-145.

[35] Maragkoudakis, P. A., Zoumpopoulou, G., Miaris, C., Kalantzopoulos, G., Pot, B., & Tsakalidou, E. (2006). Probiotic potential of *Lactobacillus* strains isolated from dairy products. International Dairy Journal, 16(3), 189-199.

[36] da Silva, J. M., Klososki, S. J., Silva, R., Raices, R. S. L., Silva, M. C., Freitas, M. Q., Barão, C. E., & Pimentel, T. C. (2020). Passion fruit-flavored ice cream processed with water-soluble extract of rice by-product: What is the impact of the addition of different prebiotic components? LWT-Food Science and Technology, 128, p. 109472.

[37] Pimentel, T. C., Madrona, G. S., & Prudencio, S. H. (2015). Probiotic clarified apple juice with oligofructose or sucralose as sugar substitutes: Sensory profile and acceptability. LWT-Food Science and Technology, 62(1), 838-846.

[38] da Costa, G. M., de Carvalho Silva, J. V., Mingotti, J. D., Barão, C. E., Klososki, S. J., & Pimentel, T. C. (2017). Effect of ascorbic acid or oligofructose supplementation on *L. paracasei* viability, physicochemical characteristics and acceptance of probiotic orange juice. LWT-Food Science and Technology, 75, 195-201.

[39] Santos, M. A., Costa, G. M., Dias, S. S., Klososki, S. J., Barão, C. E., Gomes, R. G., & Pimentel, T. C. (2019). Pasteurised sugarcane juice supplemented with *Lactobacillus casei* and prebiotics: physicochemical stability, sensory acceptance and probiotic survival. International Food Research Journal, 26(4), 1315-1325.

[40] de Oliveira, M. A., de Souza, V. M., Bergamini, A. M. M., & de Martinis, E. C. P. (2011). Microbiological quality of ready-to-eat minimally processed vegetables consumed in Brazil. Food Control, 22, 1400-1403.

[41] Soccol, C. R., Vandenberghe, L. P. S., Spier, M. R., Medeiros, A. B. P., Yamaguishi, C. T., Lindner, J. D., et al. (2010). The potential of probiotics. Food Technology Biotechnology, 48, 413-434.

[42] Pereira, A. L. F., Maciel, T. C., & Rodrigues, S. (2011). Probiotic beverage from cashew apple juice fermented with *Lactobacillus casei*. Food Research International, 44(5), 1276-1283.

[43] Leite, A. K., Santos, B. N., Fonteles, T. V., & Rodrigues, S. (2021). Cashew apple juice containing gluco-oligosaccharides, dextran, and tagatose promotes probiotic microbial growth. Food Bioscience, 42, p. 101080.

[44] Silva, S. B., & Ferrari, J. (2016). Development of Probiotic Grape Juice and Lactobacillus paracasei Viability under Cold Storage. In X CIGR Section IV International Technical Symposium, XXV Congresso Brasileiro de Ciência e Tecnologia de Alimentos.

[45] Malganji, S., Sohrabvandi, S., Jahadi, M., Nematollahi, A., & Sarmadi, B. (2016). Effect of refrigerated storage on sensory properties and viability of probiotic in grape drink. Applied Food Biotechnology, 3(1), 59-62.

[46] Yuasa, M., Shimada, A., Matsuzaki, A., Eguchi, A., & Tominaga, M. (2021). Chemical composition and sensory properties of fermented citrus juice using probiotic lactic acid bacteria. Food Bioscience, 39, p. 100810.

[47] de Oliveira Vieira, K. C., Ferreira, C. D. S., Bueno, E. B. T., De Moraes, Y. A., Toledo, A. C. C. G., Nakagaki, W. R., Pereira, V. C., & Winkelstroter, L. K. (2020). Development and viability of probiotic orange juice supplemented by *Pediococcus acidilactici* CE51. LWT-Food Science and Technology, *130*, p. 109637.

[48] Sengun, I. Y., Kirmizigul, A., Atlama, K., & Yilmaz, B. (2020). The viability of *Lactobacillus rhamnosus* in orange juice fortified with nettle (*Urtica dioica* L.) and bioactive properties of the juice during storage. LWT-Food Science and Technology, 118, p. 108707.

[49] Miranda, R. F., de Paula, M. M., da Costa, G. M., Barão, C. E., da Silva, A. C. R., Raices, R. S. L., Gomes, R. G., & Pimentel, T. C. (2019). Orange juice added with *L. casei*: is there an impact of the probiotic addition methodology on the quality parameters? LWT-Food Science and Technology, 106, 186-193.

[50] AdebayoTayo, B., & Akpeji, S. (2016). Probiotic viability, physicochemical and sensory properties of probiotic pineapple juice. Fermentation, 2(4), 20.

[51] Nguyen, B. T., Bujna, E., Fekete, N., Tran, A., Rezessy-Szabo, J. M., Prasad, R., & Nguyen, Q. D. (2019). Probiotic beverage from pineapple juice fermented with *Lactobacillus* and *Bifidobacterium* strains. Frontiers

in Nutrition, 6, 54.

[52] Ryan, J., Hutchings, S. C., Fang, Z., Bandara, N., Gamlath, S., Ajlouni, S., & Ranadheera, C. S. (2019). Microbial, physico chemical and sensory characteristics of mango juice enriched probiotic dairy drinks. International Journal of Dairy Technology, 73(1), 182-190.

[53] Reddy, L. V., Min, J. H., & Wee, Y. J. (2015). Production of probiotic mango juice by fermentation of lactic acid bacteria. Microbiology and Biotechnology Letters, 43(2), 120-125.

[54] Wang, J., Xie, B., & Sun, Z. (2021). Quality parameters and bioactive compound bioaccessibility changes in probiotics fermented mango juice using ultraviolet-assisted ultrasonic pre-treatment during cold storage. LWT-Food Science and Technology, 137, p. 110438.

[55] Mousavi, Z. E., Mousavi, S. M., Razavi, S. H., Emam-Djomeh, Z., & Kiani, H. (2011). Fermentation of pomegranate juice by probiotic lactic acid bacteria. World Journal of Microbiology and Biotechnology, 27(1), 123-128.

[56] Mantzourani, I., Kazakos, S., Terpou, A., Alexopoulos, A., Bezirtzoglou, E., Bekatorou, A., & Plessas, S. (2019). Potential of the probiotic *Lactobacillus plantarum* ATCC 14917 strain to produce functional fermented pomegranate juice. Foods, 8(1), 4.

[57] Oruç, S. Ö., & Çakır, İ. (2019). Probiyotik kültürlerle fermente karpuz suyu üretimi üzerine bir araştırma. Gıda, 44(6), 1030-1041.

[58] Fonteles, T. V., Costa, M. G. M., de Jesus, A. L. T., Fontes, C. P. M. L., Fernandes, F. A. N., & Rodrigues, S. (2013). Stability and quality parameters of probiotic cantaloupe melon juice produced with sonicated juice. Food and Bioprocess Technology, 6(10), 2860-2869.

[59] Mantzourani, I., Nouska, C., Terpou, A., Alexopoulos, A., Bezirtzoglou, E., Panayiotidis, M. I., Galanis, A., & Plessas, S. (2018). Production of a novel functional fruit beverage consisting of cornelian cherry juice and probiotic bacteria. Antioxidants, 7(11), 163.

[60] Wu, Y., Li, S., Tao, Y., Li, D., Han, Y., Show, P. L., Wen, G., & Zhou, J. (2021). Fermentation of blueberry and blackberry juices using *Lactobacillus plantarum*, *Streptococcus thermophilus* and *Bifidobacterium bifidum*: Growth of probiotics, metabolism of phenolics, antioxidant capacity

in vitro and sensory evaluation. Food Chemistry, 348, p. 129083.

[61] Zhang, Y., Liu, W., Wei, Z., Yin, B., Man, C., & Jiang, Y. (2021). Enhancement of functional characteristics of blueberry juice fermented by *Lactobacillus plantarum*. LWT-Food Science and Technology, 139, p. 110590.

[62] Panda, S. K., Behera, S. K., Qaku, X. W., Sekar, S., Ndinteh, D. T., Nanjundaswamy, H. M., Ray, R.C & Kayitesi, E. (2017). Quality enhancement of prickly pears (Opuntia sp.) juice through probiotic fermentation using *Lactobacillus fermentum*-ATCC 9338. LWT-Food Science and Technology, 75, 453-459.

[63] Pakbin, B., Razavi, S. H., Mahmoudi, R., & Gajarbeygi, P. (2014). Producing probiotic peach juice. Biotechnology and Health Sciences, 1(3), e24683.

[64] Wang, C. Y., Ng, C. C., Su, H., Tzeng, W. S., & Shyu, Y. T. (2009). Probiotic potential of noni juice fermented with lactic acid bacteria and bifidobacteria. International Journal of Food Sciences and Nutrition, 60(6), 98-106.

[65] Zandi, M. M., Hashemiravan, M., & Berenjy, S. (2016). Production of probiotic fermented mixture of carrot, beet and apple juices. Journal of Paramedical Sciences, 7(3), 17-23.

[66] Pereira, A. L. F., Feitosa, W. S. C., Abreu, V. K. G., de Oliveira Lemos, T., Gomes, W. F., Narain, N., & Rodrigues, S. (2017). Impact of fermentation conditions on the quality and sensory properties of a probiotic cupuassu (*Theobroma grandiflorum*) beverage. Food Research International, 100, 603-611.

GARLIC (*Allium sativum* L.) AS FUNCTIONAL AND MEDICINAL FOOD

Asma HARB[1], Banu Yeşim BÜYÜKAKINCI[2]*

[1] Istanbul Aydın University, Faculty of Engineering, Department of Food Engineering, 34295, Istanbul, Türkiye
[2] Haliç University, Faculty of Fine Arts, Department of Textile and Fashion Design, 34060, Istanbul, Türkiye
**Corresponding Author: banuyesimbuyukakinci@halic.edu.tr*
ORCID number: 0000-0001-7597-4406

ABSTRACT

Garlic (*Allium sativum L.*), a bulb herbal plant had a history of several thousand years of human consumption and use, starting as a seasoning, traditional treatment, and functional food. Nowadays, scientific studies considered this food an excellent health-promoting element. All that is because of the existence of active organosulfur compounds (OSCs) in garlic, which are disease-preventing for many common diseases, from cancer, heart, and blood vessel disorders, metabolic disturbances, and blood pressure to diabetes mellitus through its antioxidant or inflammation treatment. Garlic as well fights several viral infections (e.g., coronavirus) and has fat-reducing properties, as proven in several in vitro, in vivo, and clinical studies. In this article, the importance of garlic & its active sulfur compounds was investigated, according to previous studies. Also, the health benefits of garlic were discussed as well.

Keywords: *Garlic (Allium sativum L.), sulfur-containing compounds, bioactive components, health benefits, anticancer mechanism.*

INTRODUCTION

Garlic (*Allium sativum L.*) is a bulb flowering plant with a history of several thousand years of human consumption and use, starting as a seasoning, traditional treatment, and a portion of functional food **[1]**. The Codex Ebers, one of the oldest information sources for medical texts mentioned garlic as therapy and included it in a lot of medication recipes. In old traditional Asian medicine, garlic has been prescribed to help patients with

[1]Geliş Tarihi: 19.07.2021 Kabul Tarihi: 22.08.2021
DOI: 10.17932/IAU.IJFER.2015.003/ijfer_v07i2003

breathing and digestive problems. Several documents highlight garlic as a medicine with magnificent healing effectiveness **[1]**. The consuming part can be the root, leaves, or bulb itself. It can also be fresh green, dried, or in the form of a powdered spice or its extract oil. All examined parts contain different types of active ingredients such as sulfur moieties such as γ-glutamyl-S-alkenyl-L-cysteine, S- alkenyl-L-cysteine sulphoxides, alliin, and ajoene **[2].** The main sulfur compound is alliin. On average, during the consumption of garlic alliinase is released and converted into allicin which is unstable and rapidly deconstructs into other products, including dithiins, ajoene, allyl methyl trisulfide, diallyl disulfide (DADS), diallyl sulfide (DAS), diallyl trisulfide (DATS) & oil-soluble organosulfur compounds (OSCs) **[3].** These OSCs are generally considered the main bioactive component in cancer prevention. S-Allylcysteine (SAC) is a stable compound known because of its antioxidant and cholesterol-reducing effects via clinical research & pharmacological action of allicin or diallyl disulfide **[1, 2].**

The mechanism of action of garlic is wide branched. The therapeutic results of garlic are remarkable. The presence of some biologically active compounds, for example, organic sulfides, saponins, and phenolic compounds **[4].** Several in vitro and in vivo experiments have shown that these compounds can modulate various signaling pathways, improving the network shape of biological activities: anti-inflammatory, antioxidant properties, chemo-preventive, antiproliferative, anti-angiogenic, antidiabetic, immunomodulatory, anti-obesity, antibacterial properties and cardioprotective effects **[2].** In Egypt, garlic's abilities as a chemotherapeutic agent against tumors have been 3500 years ago **[5]**. A lot of food processing approaches were working on manufacturing techniques to decrease the loose of the functionality of garlic's major compounds **[6].** Various manufacturing techniques (during dehydration, chopping, and packaging) were discussed in related articles to play an important role in garlic preservation. However, their only object is not to decrease garlic quality. The findings of this review can provide a scientific basis to help improve the methods of handling garlic & its multiform products **[7]**.

This article represents a summary of studies that investigated the roles of garlic as a functional and medicinal food. Also, discusses the promise of using garlic's bioactive compounds can lead to improvement between natural food therapy and pharmacological drugs.

Table 1. The chemical combination of garlic bulbs **[8,9]**.

Chemical composition	Percent %
H_2O	65%
Carbohydrate	28%
Protein	2%
Amino acid Fiber, fatty acid, phenols, trace elements	1.2%
Sulfur-containing compounds	1.5%
Vitamins (C, B) & minerals (K, Ca, Se).	2.3%

Chemistry of garlic

The chemical combination of garlic bulbs contains different materials shown in Table 1. While the active main organosulfur components of garlic (Figure 1) like diallyl thiosulfonate (allicin), diallyl sulfide (DAS), diallyl disulfide (DADS), diallyl trisulfide (DATS), E/Z-ajoene, S-allyl-cysteine (SAC) and S-allyl-cysteine sulfoxide (alliin). Alliin or S-allyl-cysteine sulfoxide is the responsible compound for flavor and taste **[4, 9]**. During the consumption of garlic, alliinase is released and converted into allicin when it enters the body to form bi-products **[3]**. Earlier studies confirmed the health-promoting properties of garlic for the human body. The health benefits are reportedly due to the presence of mentioned bioactive compounds **[9, 10]**.

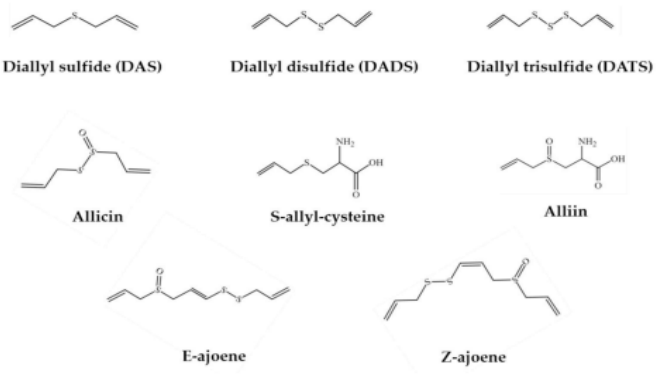

Figure 1. The chemical structural form of the main organosulfur compounds in garlic **[10]**.

In general, these bioactive compounds vary in their bioavailability and digestibility and their levels can vary depending on whether the garlic is fresh, cooked, dried, or chopped **[4]**. In a randomized controlled study aims to determine the residues of garlic components in the body, after oral consuming 1-3 grams of fresh garlic, organosulfur compound (diallyl disulfide - DADS and diallyl sulfide - DAS) was not detected in urine samples after 6-24 hours. while compound like allyl methyl sulfide which can be detected in Breath undergoes extensive metabolism produced from allyl thiosulfate **[11].** The effects of different processing methods and storage conditions (humidity, temperature, pH) along with the efficiency of bioactive components have been also included **[12]**. In the botany field, the defense mechanisms of garlic against plant pests, and pathogens have also been shown in laboratory experiments to prevent cell damage and plant tissue lesions **[13].**

Biological Activities of Garlic

Antioxidant activity

On the cellular level, the last effect of active ingredients such as phenols and saponins appear as major antioxidant compounds **[4, 14, 15].** While the intensity of this effect and the mechanism of action appear as a network full of branches (pathway regulation) and side products (minor products) **[9].**

Anti-inflammatory activity

The bioactivity of garlic has been shown to exhibit anti-inflammatory properties **[10].** Moreover, allicin could be included in therapy as an anti-inflammatory against arthritis disease, due to its low presence of toxicity. **[16].** A study by Martina et al focused on the mechanism of the ethyl linoleate which controlled the number of oxidative chemicals (nitric oxide -NO and prostaglandin E-2) by decreasing regulation & expression through their pathway either by inhibiting the transcription factor or enhancing the macrophage's effectiveness **[4, 14].** Also, garlic supplements consider to stopping inflammatory agents, which can be nitric oxide (NO), tumor necrosis factor α (TNF-α), or interleukin-1 (IL-1) according to pharmacological research **[16].**

Antimicrobial activity

During the research on the antibacterial and antifungal properties of garlic in Italy, it was found that garlic significantly suppresses the growth of *Aspergillus Versicolor*, *Penicillium citrinum,* and *Penicillium expansum*. Also, the oil extracted from the fresh plant got the ability to prevent the production of toxins from bacteria and may limit the growth of *Staphylococcus aureus*, *Escherichia coli,* and *Bacillus subtilis* by destroying the formation and the metabolic pathway of bacterial cells **[9, 14]**. Furthermore, in a clinical trial, the treatment of raw garlic inhibited *Helicobacter pylori* in the stomach of patients with *H. pylori* infection **[13]**.

Garlic protective effect

During eating products containing garlic, organosulfur compounds (e.g., allicin and alliin) and flavonoid (e.g., quercetin) will be responsible for most immunomodulatory effects. Treatment with garlic oil for 30 minutes can normalize several immunological factors from serum total immunoglobulin concentration to T-cell subtype CD4+. Moreover, consuming AGE reduces the severity of influenza by improving body defense responses **[14, 17, 18].**

In another study, dried garlic can effectively reduce blood total fat, and inhibit platelet aggregation and any risk factors along with heart diseases. All by increasing the production of NO and hydrogen sulfide (H_2S) and inhibiting the angiotensin-converting enzyme, thereby lowering hypertension and oxidative stress **[10, 16].** The anti-hypertension mechanisms of garlic are shown in (Figure 2). The findings of randomized controlled research, have suggested that garlic supplementation can reduce atherosclerosis or prevent cardiovascular events, such as myocardial infarction or stroke but the exact technique is still not fully understood **[9, 19]**

In the elderly category, ingredients such as S-allyl mercaptocysteine stood out. Where they help activate the role of antioxidants in the body. Which was used in the composition of medicines given to Alzheimer's patients because of its association with nerve impulses and brain function **[17, 20]**. The sector of garlic as a hypoglycemic agent is based either on stimulation of the pancreas that increases the secretion of insulin from β-cells and increases insulin sensitivity or the release of bound insulin which controls

the glucose content in the blood. Vitamin B6 and Vitamin C are present in garlic extract. Vit B is engaged in the metabolism of carbohydrates, while Vit. C plays an important role in controlling and maintaining the sugar level in the blood **[21]**. Garlic oil, obtained from steam distillation of Allium sativum enhances insulin and glucose tolerance. It also enhances skeletal muscle glycogenesis **[15, 22]**. Allicin in combination with sulfur-containing amino acid, will create a pathway involved in the regulation and increase the blood insulin concentration by preventing the inactivation of insulin by the liver. Garlic functions as an insulin secretagogue or insulinotropic in diabetic rats **[2, 3, 21]**. A study by Sudhakar et al., (2021) demonstrated in Table 2 used supplements containing garlic in the management of type 2 diabetes mellitus and reducing diabetic complications **[21].**

Anticancer mechanisms of garlic

The anticancer-related study has shown that allylsulfide derivatives have significant and clear biological effects on mechanisms in carcinogenesis **[19].** The anticancer actions have been divided into the following categories depending on the cancer phase, which are summarized in Figure 3.

Table 2. Summary of clinical trials of herbal medicine in diabetic patients **[21].**

TYPE OF FOOD	MECHANISM	CLINICAL TRIALS	AGE GROUP (YEARS)	OUT-COMES	COM-PLI-CAT-ION
Allium sativum (Garlic tablet)	Allicin combines with cysteine and enhances serum insulin. Inhibiting the forming of advanced glycation final products (AGEs) It increases the pancreatic secretion of insulin from the beta cells.	Type 2 Diabetes Mellitus patients	25-70 years	Improve the glycemic control by improving the glycemic index and maintaining a glucose level	Gastric irritation
	Direct hypoglycemic action, fasting blood glucose level and serum fructosamine are decreased significantly.	Type 2 diabetes atherosclerosis patients	34-62 years	Fast blood glucose level was decreasing, and serum fructosamine was declining.	Not stated

In the initiation phase of cancer progression, it inhibits vital phytochemicals and carcinogens with antioxidants, mutagenicity, and detoxification. In the second phase (promotion), it cuts the reproduction cycle of phytochemicals that lead to stopping the propagation of cells by folding protein and DNA repairing. In the third phase (progression), the inhibition of phytochemicals causes changing cell pathways, including antiproliferation, apoptosis, and immunocompetence **[19].**

Previous in vitro studies have used the extraction material from garlic in inhibiting mutagenicity in bacteria, which came very effectively by SAC and SAMC the strong radical activities that block the extracellular mutagens **[19].** On the other hand, the consumption of garlic increased circulatory antioxidants, vitamin E, and superoxide dismutase. While reducing glutathione, glutathione peroxidase, and peroxidation of lipids **[23, 24].** Stimulating scavenging of reactive oxygen species (ROS) prevents oxidative damage to healthy tissue. The enzymatic activity of cytochrome P450 s (CYP2E1) was affected by the DAS oxidant derivative, leading to stop the production of carbon tetrachloride, acetaminophen, and N-nitroso dimethylamine which consider toxic carcinogen-forming elements **[23, 24, 25].** Additionally, the ajoene component causes an accumulation and activation of misfolded protein aggregates in cancer cells **[19, 23].** For all that, garlic's functional components consider blockers and regulators important in the first and second stages of oncogenesis. Prevent transportation of the tumor between organs by inhibiting toxic chemicals or interference between cellular macromolecules in DNA, RNA, and proteins **[3, 24].** When cancer is invasive and metastatic, even immune evasion, growth, and proliferation can be happening. Garlic here is involved in the induction of apoptosis (death) of damaged cells by an increased splitting of genetic material and intracellular Ca, p53, Bax, and down-regulation Bcl-2.

A novel study has incubated One hundred and eighty cancer cells or what is known as Murphy-Sturm lymphosarcoma cells with diethyl thiosulfate. After that, it revealed that a complete prevention effect of cancer cells from developing tumors was noticed **[2, 23, 24]**. In vitro level, recent studies represent an idea that *Allium sativum* (garlic) has antiproliferative characteristics and apoptotic activities on carcinoma cell line A498 located in the kidney and carcinoma cell line A549 cell lines in the lung while no effect of a toxic level was detected **[3, 9]**. At the same level, the combination

of S-ally cysteine from *Allium sativum* and lycopene from tomatoes can form suppress chemically induced gastric cancer progression **[24, 19]**. The functional components of garlic are not only effective in the chemopreventive stages of cancer but also contribute to enhancing the effectiveness of cancer treatment and reducing the side effects of treatment, whether chemo or radiotherapy, by suppressing symptoms after sessions. This result is quite surprising in the difference in the therapeutic effect of taking garlic extraction. Recent information indicated that the phytochemicals of raw garlic have a very high selectivity to kill exceptionally efficient cancer cells with no damage to normal cells **[19]**.

Against coronavirus

Seventeen active organosulfur compounds are found in garlic essential oil, results show that 99.4% of contents have a strong effect on basic units of the ACE2 protein and PDB6LU7 protease of SARS-CoV-2. Also, it considers that these essential oils are a valuable natural antivirus source and can help prevent coronavirus invasion into the human body **[26, 27].** Since the scientific research race against the coronavirus began, many drugs have been produced, either to fight the virus itself or the side effects and even some research has worked to produce drugs that help stimulate the human immune system **[27]**.

Therapeutic Promises of Garlic

Medical importance garlic is an excellent source of therapeutic agents. A pharmaceutical drug from garlic was produced that contains allicin organosulfur compounds. For better results, garlic should be fresh and chemical-free. Pure allicin has been found effective against fungus and mold accumulation. The garlic was smashed to be used for sores in the mouth and throat, while these days it can be used in toothpaste to prevent related problems **[2]**. It is also good for hair growth (since it consists of vitamin B-6 and Vit C which are good for hair growth, nowadays a lot of shampoos and hair treatment products contain sulfur as an additive from garlic), for flu, anti-febrile and Anti-helminthic activity **[3, 28].**

Garlic also helps in removing the *Giardia lamblia* parasite from the intestinal tract. Antimicrobial activity: the allicin consists of organosulfur compounds which are the most important for antimicrobial properties **[13, 28].** These findings, if used in the right ways, may give a new valuable method for treating cancer using traditional medicinal plants **[2, 29, 30].**

CONCLUSION

Garlic (*Allium sativum L*.) is a herbal plant used worldwide and considered a food, seasoning and old cultural (medicinal) treatment for the prevention of infectious diseases since antiquity. Through research, the food sciences and pharmaceutical industries have developed links between garlic's functional effect on cancer, heart disorders, blood pressure, and diabetes mellitus. Antioxidants and anti-inflammatories that fight many viral infections and have lipid-lowering properties have also been studied, as shown in numerous research and clinical studies. This article summarizes scientific investigations into the powerful effects of garlic and its wide use as a functional and medicinal food.

REFERENCES

[1] Wu, Q., & Liang, X. (2018). Food therapy and medical diet therapy of Traditional Chinese Medicine. Clinical Nutrition Experimental, 18, 1-5.

[2] Ashfaq, F., Ali, Q., Haider, M. A., Hafeez, M. M., & Malik, A. (2021). Therapeutic activities of garlic constituent phytochemicals. Biological and Clinical Sciences Research Journal, 2021(1).

[3] Ansary, J., Forbes-Hernández, T. Y., Gil, E., Cianciosi, D., Zhang, J., Elexpuru-Zabaleta, M., Simal-Gandara, J., Giampieri, F., & Battino, M. (2020). Potential health benefit of garlic based on human intervention studies: A brief overview. Antioxidants, 9(7), 619.

[4] Martins, N., Petropoulos, S., & Ferreira, I. C. (2016). Chemical composition and bioactive compounds of garlic (*Allium sativum* L.) as affected by pre-and post-harvest conditions: A review. Food Chemistry, 211, 41-50.

[5] Majewski, M. (2014). *Allium sativum*: facts and myths regarding human health. Roczniki Państwowego Zakładu Higieny, 65(1).

[6] De Greef, D., Barton, E. M., Sandberg, E. N., Croley, C. R., Pumarol, J., Wong, T. L., Das, N., & Bishayee, A. (2021). Anticancer potential of garlic and its bioactive constituents: a systematic and comprehensive review. In Seminars in cancer biology (Vol. 73, pp. 219-264). Academic Press.

[7] Feng, Y., Xu, B., Yagoub, A. E. A., Ma, H., Sun, Y., Xu, X., Yu, X., & Zhou, C. (2021). Role of drying techniques on physical, rehydration, fla-

vor, bioactive compounds, and antioxidant characteristics of garlic. Food Chemistry, 343, 128404.

[8] Tavares, L., Santos, L., & Noreña, C. P. Z. (2021). Bioactive compounds of garlic: A comprehensive review of encapsulation technologies, characterization of the encapsulated garlic compounds, and their industrial applicability. Trends in Food Science & Technology, 114, 232-244.

[9] Shang, A., Cao, S. Y., Xu, X. Y., Gan, R. Y., Tang, G. Y., Corke, H., Mavumengwana, V., & Li, H. B. (2019). Bioactive compounds and biological functions of garlic (*Allium sativum* L.). Foods, 8(7), 246.

[10] Morales-González, J. A., Madrigal-Bujaidar, E., Sánchez-Gutiérrez, M., Izquierdo-Vega, J. A., Valadez-Vega, M. D. C., Álvarez-González, I. Morales-González, Á., & Madrigal-Santillán, E. (2019). Garlic (*Allium sativum* L.): A brief review of its antigenotoxic effects. Foods, 8(8), 343.

[11] Mondal, S., Soumya, N. P. P., Mini, S., & Sivan, S. K. (2021). Bioactive compounds in functional food and their role as therapeutics. Bioactive Compounds in Health and Disease, 4(3), 24-39.

[12] Li, G., Chen, P., Zhao, Y., Zeng, Q., Ou, S., Zhang, Y., Wang, P., Chen, N., & Ou, J. (2021). Isolation, structural characterization, and antioxidant activity of a novel polysaccharide from garlic bolt. Carbohydrate Polymers, 267, 118194.

[13] Bhatwalkar, S. B., Mondal, R., Krishna, S., Adam, J. K., Govender, P., & Anupam, R. (2021). Antibacterial Properties of Organosulfur Compounds of Garlic (*Allium sativum*). Frontiers in microbiology, 12, 613077.

[14] Kopeć, A., Skoczylas, J., Jędrszczyk, E., Francik, R., Bystrowska, B., & Zawistowski, J. (2020). Chemical composition and concentration of bioactive compounds in garlic cultivated from air bulbils. Agriculture, 10(2), 40.

[15] Melguizo-Rodríguez, L., García-Recio, E., Ruiz, C., De Luna-Bertos, E., Illescas-Montes, R., & Costela-Ruiz, V. J. (2022). Biological properties and therapeutic applications of garlic and its components. Food & Function.

[16] Sasi, M., Kumar, S., Kumar, M., Thapa, S., Prajapati, U., Tak, Y... & Mekhemar, M. (2021). Garlic (*Allium sativum* L.) Bioactives and their role in alleviating oral pathologies. Antioxidants, 10(11), 1847.

[17] Agahi, F., Penalva-Olcina, R., Font, G., Juan-García, A., & Juan, C. (2022). Effects of Voghiera garlic extracts in neuronal human cell line against zearalenone's derivates and beauvericin. Food and Chemical Toxicology, 112905.

[18] Samtiya, M., Aluko, R. E., Dhewa, T., & Moreno-Rojas, J. M. (2021). Potential health benefits of plant food-derived bioactive components: An Overview. Foods, 10(4), 839.

[19] Farhat, Z., Hershberger, P. A., Freudenheim, J. L., Mammen, M. J., Hageman Blair, R., Aga, D. S., & Mu, L. (2021). Types of garlic and their anticancer and antioxidant activity: A review of the epidemiologic and experimental evidence. European Journal of Nutrition, 60(7), 3585-3609.

[20] Wilson, D. W., Nash, P., Buttar, H. S., Griffiths, K., Singh, R., De Meester, F., Horiuchi, R., & Takahashi, T. (2017). The role of food antioxidants, benefits of functional foods, and influence of feeding habits on the health of the older person: An overview. Antioxidants, 6(4), 81.

[21] Sudhakar, K., Mishra, V., Hemani, V., Verma, A., Jain, A., Jain, S., & Charyulu, R. N. (2021). Reverse pharmacology of phytoconstituents of food and plant in the management of diabetes: Current status and perspectives. Trends in Food Science & Technology, 110, 594-610.

[22] Panyod, S., Wu, W. K., Chen, P. C., Chong, K. V., Yang, Y. T., Chuang, H. L., ... & Sheen, L. Y. (2022). Atherosclerosis amelioration by allicin in raw garlic through gut microbiota and trimethylamine-N-oxide modulation. NPJ Biofilms and Microbiomes, 8(1), 1-13.

[23] Özkan, İ., Koçak, P., Yıldırım, M., Ünsal, N., Yılmaz, H., Telci, D., & Şahin, F. (2021). Garlic (*Allium sativum*)-derived SEVs inhibit cancer cell proliferation and induce caspase mediated apoptosis. Scientific reports, 11(1), 1-11.

[24] Aghajanpour, M., Nazer, M. R., Obeidavi, Z., Akbari, M., Ezati, P., & Kor, N. M. (2017). Functional foods and their role in cancer prevention and health promotion: a comprehensive review. American Journal of Cancer Research, 7(4), 740.

[25] Zhang, Y., Liu, X., Ruan, J., Zhuang, X., Zhang, X., & Li, Z. (2020). Phytochemicals of garlic: Promising candidates for cancer therapy. Biomedicine & Pharmacotherapy, 123, 109730.

[26] Khubber, S., Hashemifesharaki, R., Mohammadi, M., & Gharibzahedi, S. M. T. (2020). Garlic (*Allium sativum* L.): a potentially unique therapeutic food rich in organosulfur and flavonoid compounds to fight COVID-19. Nutrition Journal, 19(1), 1-3.

[27] Thuy, B. T. P., My, T. T. A., Hai, N. T. T., Hieu, L. T., Hoa, T. T., Thi Phuong Loan, H., ... & Nhung, N. T. A. (2020). Investigation into SARS-CoV-2 resistance of compounds in garlic essential oil. ACS Omega, 5(14), 8312-8320.

[28] Rouf, R., Uddin, S. J., Sarker, D. K., Islam, M. T., Ali, E. S., Shilpi, J. A., Nahar, L., Tiralongo, E., & Sarker, S. D. (2020). Anti-viral potential of garlic (*Allium sativum*) and its organosulfur compounds: a systematic update of pre-clinical and clinical data. Trends in food science & technology.

[29] Li, S., Li, N., Wang, Y., Wang, Q., Li, R., Zhang, M., Panichayupakaranant, P., Gao, Y., & Chen, H. (2022). Structural, functional, and proteomic differences of proteins extracted from white garlic and Laba garlic. Food Research International, 155 111047.

[30] Moreno-Ortega, A., Di Pede, G., Pereira-Caro, G., Calani, L., Mena, P., Del Rio, D., & Moreno-Rojas, J. M. (2022). In vitro colonic fermentation of (poly) phenols and organosulfur compounds of fresh and black garlic. Journal of Agricultural and Food Chemistry, 70(12), 3666-3677.